IN THE UNITED STATES DISTRICT COURT
FOR THE NORTHERN DISTRICT OF OKLAHOMA

STATE OF OKLAHOMA, ex rel. W.A. DREW EDMONDSON, in his capacity as ATTORNEY GENERAL OF THE STATE OF OKLAHOMA AND OKLAHOMA SECRETARY OF THE ENVIRONMENT C. MILES TOLBERT, in his capacity as the TRUSTEE FOR NATURAL RESOURCES FOR THE STATE OF OKLAHOMA **PLAINTIFFS**

v. **CASE NO.: 05-CV-00329 GKF –SAJ**

TYSON FOODS, INC., TYSON POULTRY, INC., TYSON CHICKEN, INC., COBB-VANTRESS, INC., CAL-MAINE FOODS, INC., CAL-MAINE FARMS, INC. CARGILL, INC., CARGILL TURKEY PRODUCTION, LLC, GEORGE'S, INC., GEORGE'S FARMS, INC., PETERSON FARMS, INC., SIMMONS FOODS, INC. and WILLOW BROOK FOODS, INC. **DEFENDANTS**

Declaration of Dr. Mansour Samadpour and Dr. Samuel P. Myoda

1. Dr. Mansour Samadpour is President of IEH Laboratories and Consulting Group and Molecular Epidemiology, Inc. (IEH). Dr. Samuel Myoda is Vice President of IEH. Our CVs are attached hereto as Exhibit 1 and 2. In the last four years, Dr. Samadpour has testified in the following cases:

   Novak v. Kroger Co. of Michigan, 02-038264-NP (Mich. Cir. Ct.)

   Bar-S Foods v. Tiromat, et al., CJ-02-111 (Okl. Dist. Ct.)

   Metz v. Dalesio's of Lilttle Italy, 24-C-06-001426OT (Baltimore Cir. Ct.)

2. IEH was retained in November of 2007 by Defendants at an hourly rate of $400/hr for Dr. Samadpour and $350 for Dr. Myoda to serve as expert consultants regarding, but not limited to, microbial source tracking, microbiology, water quality, public health and related regulatory affairs. We have been asked to review the Plaintiff's testimony, the water quality data and other relevant parameters that influence water quality in the Illinois River Watershed (IRW) and to offer our scientific opinions on the water quality and factors that affect the water quality in the IRW.

3. Bacteria are microscopic, unicellular organisms that are prokaryotes, meaning that unlike our cells they do not contain a nucleus. There are many different physical and biological properties of these organisms, they have preferred habitats, and interact with their surroundings in many different ways. There are literally trillions upon trillions of bacteria in the environment and the overwhelming majority of them are not pathogenic. Bacteria are an integral part of our lives; they are used in food production, e.g. yogurt and cheese, in our digestive system, e.g. *E. coli* and enterococcus, in the soil, e.g. nitrogen fixing bacteria that are essential to the nutrient cycle and are also in the air and water. Bacteria are used to treat wastewater, to break down pollutants, e.g. bioremediation, and are used in drug development and production. There are a plethora of beneficial uses for bacteria.

4. Despite the wealth of knowledge regarding bacteria, in fact relatively little is known about the total universe of bacteria, and it is estimated that scientists have been able to culture (grow in the lab) < 2% of the bacteria that exist. There are a host of factors that affect the "fate and transport" of specific types of bacteria in the environment: how do different bacteria handle different environmental factors; what is the

relationship between bacterial transport and precipitation; what factors govern soil infiltration and filtration/sorption; what characteristics govern relative survival rates, predation rates, and growth rates; and the impact of factors such sunlight (UV radiation), temperature, sedimentation, humidity, pH, moisture content, etc.? These factors affect the fate and transport of each bacterium.

5. Developing strategies to protect public health has driven the study of microbiology. In the late 1800s/early 1900s, illness due to poor sanitation and water quality was commonplace, largely due to inadequate sewage treatment, hence the need to develop and install sewage treatment facilities and limit, if not prevent, fecal matter from entering the water. In order to evaluate the effectiveness of the treatment interventions and resulting water quality, a test was needed. The ideal approach is to test for the pathogens in the water directly; however, many of the pathogens were unknown, in relatively small quantities and hard if not impossible to test for at the time. Therefore, the indicator organism approach was embraced and because certain coliforms are virtually always found in feces, the presence of total coliforms was used as an indication that fecal contamination was probable. The total coliform group contains bacteria such as Citrobacter, Enterobacter, Escherichia (*E. coli*), Hafnia, Klebsiella, Serratia and Yersinia.

6. The problem with using total coliforms as an indicator of fecal contamination is that not all the bacteria in this group are found in feces. In an effort to increase the accuracy of the indicator organism approach, a subset of total coliforms – the faecal coliform group (aka fecal coliform) replaced total coliforms as a measure of water quality. The faecal coliform group contains bacteria such as *E. coli*, Citrobacter,

Enterobacter and Klebsiella. Although this was a better approach, it still did not eliminate the possibility that indicators would be found when in fact there was no fecal contamination (or pathogens) present. This is because in the faecal coliform group, *E. coli* is associated with feces however; organisms such as Citrobacter, Enterobacter and Klebsiella do not originate in feces and because the faecal coliform count does not differentiate which bacteria(s) are being quantified, it is not known which organisms are present/absent or where they came from.

7. In the early 1980s, realizing the shortcomings of the use of faecal coliforms as an indicator, the EPA set out to develop a better methodology to measure water quality. The technology was still not in place to directly detect all the potential pathogens so alternative indicator bacteria were evaluated. By definition, indicator(s) must be easy to detect, non pathogenic, exist in greater quantities than the pathogen and must live longer than the pathogens. An indicator would be useless if it did not persist in the environment at least as long as the pathogen. In addition, there should be a correlation of the indicator concentration with the pathogen concentration and the fate and transport properties of the indicator and pathogen should be identical/similar. Unfortunately, the EPA did not evaluate the correlation of indicator with pathogen or the fate and transport characteristics of each. The EPA did however; evaluate the indicator concentration versus number of reported gastrointestinal illnesses reported by swimmers using the water bodies for primary contact recreation although they did not attempt to identify which pathogens were causing illnesses. The general consensus in the scientific community is that the majority of the illnesses were caused by enteric viruses; the anecdotal evidence supports this conclusion as the studies were done at beaches that were impacted by wastewater discharges that

would typically carry human enteric viruses. The conclusions of the epidemiological studies resulted in issuance of the *Ambient Water Quality Criteria for Bacteria -1986* (EPA440/5-84-002) that recommended a water quality standard of a geometric mean of 126CFU *E. coli*/100ml (235CFU/100ml to 576CFU/100ml single sample maximum)or 33 CFU enterococcus/100mL (61CFU/100ml to 151CFU/100ml single sample maximum) for fresh water and 35CFU enterococcus/100ml (104CFU/100ml to 500CFU/100ml single sample maximum) for marine water (based on the EPA determination that the acceptable illness rate is 8 to 19 illnesses per 1,000 swimmers). After its issuance, the EPA recommended that all States use either *E. coli* or enterococcus instead of faecal coliforms as indicators of water quality and in 2000, the Beaches Environmental Assessment and Coastal Health (BEACH) Act required States adjacent to the Great Lakes and coastal states to adopt the 1986 Standards.

8. The recommendation to change indicators was met with resistance due to the reluctance to change for a variety of reasons, including but not limited to the fact that the correlation of illnesses to indicator concentrations was not as strong as some deemed appropriate and that the epidemiological studies were carried out in waters that were impacted by wastewater treatment plant discharges. One of the major objections was that in areas that have wastewater treatment plant effluent the illness rate versus indicator concentration would be higher due to the presence of human enteric viruses in the effluent. At the time of the studies, the EPAs intention was to do additional studies to determine if indicators that were derived from various sources did in fact hold a different correlation with illness rates. However, the studies were never carried out due to funding constraints. The indicators, both *E. coli* and enterococcus are shed from virtually all warm blooded animals, e.g. cattle, pigs, deer,

birds, wildlife, waterfowl, humans, pets, etc. In fact, wildlife, waterfowl and birds are major contributors of the *E. coli* and enterococcus that are found in surface waters. The issue of different sources was addressed with an EPA policy that stated a State could discount all indicator bacteria derived from nonhuman sources when making regulatory decisions. This policy was extremely important because States were in the process of developing total maximum daily load regulations (TMDLs) to address high levels of bacteria in surface waters throughout the country and finding that *E. coli* and enterococcus were ubiquitous in the environment. Relative to the EPAs recommended standards, surface waters throughout the country are out of compliance. For example, Delaware a state with three counties, one primarily urban, one primarily agriculture and one mixed lists approximately 97% of the State's waters on the 303(d) list as impaired due to high indicator bacteria levels. Coincidently, Delaware monitors approximately 97% of its surface waters. In Oklahoma, 5,847 miles of stream segments are listed as impaired due to high enterococcus levels, 3,118 miles due to high *E. coli* levels and 2,921 miles due to high faecal coliform levels. Of the lakes assessed, 34% did not meet the primary contact recreation standards (Tenkiller Ferry Lake is not impaired by bacteria). In Oklahoma, more stream segments are listed as impaired for enterococcus than any other water quality parameter. In the IRW there are no stream segments listed as impaired by faecal coliforms, 8.6 miles listed as impaired by *E. coli* and 97.2 miles listed as impaired by enterococcus. This represents 0% of the States impairments for faecal coliforms, 0.28% of the States impairments for *E. coli* and only 1.78% of the States impairments for enterococcus (Oklahoma 2006 303(d) list). The issue of high bacteria levels is prevalent throughout Oklahoma and is in no way confined to the IRW or areas that are used for poultry production or the application of poultry litter. The bacteria levels that are seen in the

IRW and throughout Oklahoma are typical of the levels seen throughout the country as evident by 303(d) listings and TMDLs that require reductions of up to and in some times greater than 90% of the indicator bacteria.

9. A TMDL is the appropriate regulatory mechanism that is in place to address impaired waters. They require monitoring, identification of the sources of a pollutant, and load allocations and pollution control strategies to remediate the problem.

10. In order to discount the nonhuman sources and determine what the sources were so that appropriate pollution control strategies (PCS) and best management practices (BMPs) could be developed, the science of microbial source tracking (MST) was developed (early/mid 1990s). MST was an extension of the principles used in track down studies such as those done by the CDC during an illness outbreak. Antibiotic resistance analysis (ARA, aka ARP – antibiotic resistance pattern) was one of the more widely used MST techniques. Initial studies reported high average rate of correct classifications (ARCC) as a measure of accuracy (using various calculation techniques including the holdout method of cross validation) and suggested that an ARCC of 60% to 70% was enough for water quality managers to base decisions on (Harwood et. al, 2000, *Classification of Antibiotic Resistance Patterns of Indicator Bacteria by Discriminant Analysis: Use in Predicting the Source of Fecal Contamination in Subtropical Waters*). Encouraged by the results of initial studies and due to the time constraints relating to the development of TMDLs and other regulatory pressures, ARA was quickly embraced by many as a mainstream technology and was widely used for MST. But as more studies were undertaken and ARA as well as other MST methods were challenged by the scientific community in method comparison studies in which

known samples were given blindly to participating labs, the majority of the methods performed poorly. In both the Southern Coastal California Water Research Project (SCCWRP)/EPA study and the USGS method comparison, ARA performed very poorly and had extremely high false positive rates (39% to 100%). Dr. Harwood participated in the SCCWRP study and utilized this methodology; her lab's performance reflected this deficiency. In addition, it was determined that the use of ARCC using techniques such as the holdout method of cross validation was not an adequate measure of the accuracy of the method (Harwood et. al., 2004, *Phenotypic library-based microbial source tracking methods: Efficacy in the California collaborative study* and Stoeckel and Harwood, 2007, *Performance, Design, and Analysis in Microbial Source Tracking Studies*). In general, library-based methods performed poorly, especially those that employed the population ecology approach to fingerprint analysis. The population ecology approach matches genetic patterns, e.g. ribotyping "fingerprints" by using mathematical algorithms to estimate the similarity between two fingerprints. In reality, two fingerprints can be very similar, often differing by only one band and be from bacteria originating from different sources. One method that is much more accurate is the molecular epidemiological approach in which only identical matches are considered to originate from the same source. This approach is used by the CDC and other regulatory agencies in track down investigations such as the determination of the source of disease outbreaks. IEH uses this analytical approach and performed the best in these method comparison studies.

11. In 2004, the EPA promulgated a rule that required the states included in the 2000 BEACH Act to adopt the 1986 bacteria standards and reversed the policy of discounting nonhuman derived bacteria indicators. However, many states that were not included

in the Act were still reluctant to switch from the faecal coliform standard. There was/is still a great deal of apprehension regarding the usage of E. coli and/or enterococcus as indicators, so much so that the 2000 BEACH Act mandated the EPA to reevaluate the standards. Dr. Myoda was invited to serve in spring of 2007 as an expert in the Experts Scientific Workshop discussing the critical research and scientific needs relating to the development of a new recreational water quality standard. Issues addressed in the workshop included but were not limited to the problem that bacteria data was not available until 24 hours after a sample was taken, there was no/weak correlations between the indicator(s) and many of the pathogens and that the correlation is variable based on the source of the indicators and the fate and transport characteristics of the indicators versus the pathogens is unclear.

12. When developing the appropriate indicator(s) to use, knowledge of the fate and transport characteristics of the indicator(s) and pathogens, both individually and as they relate to each other is critical. Individually, fate and transport is significant because only those pathogens that are present and viable in the water pose a potential public health risk to those recreating in the water. As the microbiological characteristics of each pathogen are significantly different, it is highly likely that their fate and transport characteristics will vary as well.

13. The most simplistic route of transport is direct deposition, e.g. cattle defecating in streams. Once the pathogen(s) (assumed to be carried in the feces of warm blooded mammals) is excreted over or in the water, the relevant questions are how long will the pathogen be viable and available. Indirect deposition of feces introduces many more variables affecting the fate and transport of the bacteria and or pathogen. First,

the fecal properties from different mammals vary substantially. One of the primary differences (aside from pathogen and indicator density) is moisture content. Very "wet" feces is more likely than "dry" feces to introduce pathogens into the environment. After defecation, the distance from the water plays an important role as well. Driven by heavy precipitation and transported primarily via surface runoff, the organisms may be washed into the surface water by sheet flow. During this transport, they are subjected to a variety of environmental factors including but not limited to UV disinfection, predation, temperature etc. that affect the proportion that will ultimately end up in surface water in which people are recreating. In the application of poultry litter, any indicator bacteria (as well as any other bacteria contained in the feces) are subjected to the conditions in the litter for great lengths of time prior to the application of the litter as a fertilizer/soil amendment. During this time, the composting processes along with natural die off kill a substantial portion of the bacteria. What bacteria may survive until application is then subjected to the aforementioned environmental factors and only a small portion (if any) will remain viable.

14. Resuspension from sand or sediment could also play an important role. There may be a reservoir of indicator(s) that could be reintroduced into the water column. Additional, regrowth of the indicator(s) could represent a source and confound the risk assessment/prediction.

15. Ideally, the indicator(s) chosen as the surrogate for the pathogens will have the same fate and transport characteristics of the pathogens themselves. However, since this is unlikely, it is important to know and relate the characteristics that are indicator(s)

specific to the pathogens so that the measurement of the indicator can be correlated to the concentration of the viable pathogens in the water and ultimately to public health risk.

16. Lastly, when studying microbiology, it is imperative that standard methods that have been accepted by the scientific community are followed. These methods should be approved by the appropriate authority such as the EPA, Standard Methods for the Examination of Water and Wastewater, and/or AOAC, etc. This ensures that, if the tests are carried out correctly, the results are reliable and reproducible. In addition to utilizing the proper testing method, sample collection must be carried out with equal rigor and quality controls. Issues such as hold time (normally included in the standard method) must be strictly adhered to or the results are invalid, e.g. exceeding the hold time on water samples that are being analyzed for bacteria concentrations could lead to higher counts due to regrowth. Statistically valid sampling plans must be followed; sample locations and the time the samples are taken must be randomly selected. A minimum number of samples must be taken to ensure that the testing reflects an accurate picture of the whole. Positive and negative controls must be used. Unless all these elements are included in a scientific study, the results are questionable if not invalid.

17. In reviewing the data it is apparent that standard methods were not followed in the Plaintiff's testing. In approximately 60% of the water samples, the 6 hour hold time mandated by the EPA for recreational water being tested for indicator bacteria (*E. coli*, enterococcus) was violated, in many cases by one to two days; therefore, this data is unreliable. In addition, the recreation water quality standard is based on a

geometric mean of no less than 5 samples taken within 30 days, a frequency that was not maintained during the Plaintiff's study. Sampling locations were not chosen randomly. It appears that the locations that the Plaintiff's thought it most likely to find what they wanted to find were chosen. Furthermore, it appears that the timing of sample collection was not randomized either, both with respect to the time of day samples were collected and the timing relative to flow conditions. Many of the samples were taken during high flow conditions during which bacteria counts will generally be higher than average due to resuspension and runoff. Based on all of these violations of standard methodologies, we believe that the data is unreliable and is biased and skewed in favor of the Plaintiff's position.

18. The CRA report revealed egregious violations in proper sampling protocols. These violations included but were not limited to samplers walking through feces into water that they then sampled, soil borers driven through feces and into the dirt when soil samples were taken and sampling tools not being disinfected between use. A review of the edge of field sample data reflects that the mean bacteria concentration for E. coli is 4,174 CFU/100ml, for enterococcus is 14,664 CFU/100ml, and for faecal coliforms is 6,371 CFU/100ml (see attachment A). Although there were a few samples reported to have concentrations of 1,600,000 CFU/100ml, those are atypical and represent outliers in the data set. However, even those outlying values are an order of magnitude below that of sewage influent (58,000,000 CFU *E. coli*/100ml, Miyanaga et. al, 2006, *Detection of Escherichia coli in the sewage influent by fluorescent labeled T4 phage*). In our view, the atypically high values are more consistent with samples taken in close proximity to a concentrated source of indicator bacteria, e.g.

cattle feces, than with runoff samples taken from areas affected by uniformly distributed indicator bacteria such as the application of poultry litter.

19. The indicator bacteria in the waters of the IRW originate from many sources. The loading from cattle is extremely significant. Typically, cattle will excrete 15 to 35 kg of feces per day. In the summer when the majority of primary contact recreation is occurring, the initial *E. coli* concentration in the feces will be approximately 3,000,000 *CFU E. coli*/gram, however after deposition the bacteria multiply and reach levels of approximately 48,000,000 CFU *E. coli*/gram (Sinton et. al, 2007, *Survival of Indicator and Pathogenic Bacteria in Bovine Feces on Pasture).* Also in the summer months the cattle tend to congregate near and in the streams in order to cool off, increasing the possibility of direct deposition into and in close proximately of the streams. This means that each day one cow will contribute roughly 960,000,000,000 *E. coli* into the environment and with approximately 200,000 head of cattle in the IRW over 192,000,000,000,000,000 CFU *E. coli* will be introduced into the environment each day.

20. In addition to cattle, there are approximately 150,000 swine and wildlife (geese, ducks, deer, turkeys, etc) and birds that live throughout the watershed as well as the wastewater treatment plant effluent and septic system loads that are sources of indicator bacteria. Wildlife sources are a significant source of fecal material and indicator bacteria. The USGS reported that in Delaware County, Oklahoma, 45% of the *E. coli* sampled came from birds and 22% came from cattle (*Reconnaissance of the Hydrology, Water Quality, and Sources of Bacterial and Nutrient Contamination in the*

Ozark Plateaus Aquifer System and Cave Springs Branch of Honey Creek, Delaware County, Oklahoma, March 1999–March 2000).

21. The presence of indicator bacteria does not mean that pathogens are present. Those that could be present include bacteria such as salmonella, campylobacter and *E. coli* O157:H7. These pathogens are carried by a variety of hosts, e.g., *E. coli* O157:H7 are primarily found in cattle, salmonella in reptiles and poultry and campylobacter in cattle, swine and poultry. Other hosts could carry these pathogens as well. The Plaintiff contends that *E. coli* O157:H7 is shed from poultry. However, there is virtually no evidence that poultry carries *E. coli* O157:H7 and the Plaintiff never tested for or found it in the litter or in the environment. Campylobacter is an organism that grows well in the conditions typically found in the digestive system. However, it does not survive well in the environment. It will die when exposed to oxygen and will also readily dehydrate and die. The Plaintiff's edge of field (EOF) sampling revealed that there was no campylobacter running off of the fields where litter had been applied[1]. Salmonella was only reported in 3 EOF samples[1], the concentrations were very low, and the source of the salmonella was not determined.

[1] No campylobacter was found in EOF water samples. Using BioSep beads, campylobacter and salmonella was reported however, using that technology the bacteria sorb onto the beads and no concentration can be determined. In addition, the reported values were all 50/bead (with the exception of one 5/bead) which raises suspicions regarding the quantification accuracy)

22. The "biomarker" that the Plaintiff claims to be poultry specific is not specific to poultry. In the extremely limited sampling of known sources used to validate this claim, the biomarker was found to be carried by ducks, geese and cattle. When retested the cattle sample came back negative and the explanation given by Dr. Harwood was that it was most likely due to contamination in the laboratory. If contamination was occurring in the laboratory the reliability of all the test results are suspect except the duck and goose positive samples which were verified to be correct.

23. Absence of proof is not proof of absence. Only 24 cattle manure composites, 2 swine manure composites, 10 duck and 10 goose manure composites, 3 septic and 3 wwtp samples were tested to validate the specificity of the biomarker. Virtually all of the sources of the indicator bacteria in the IRW were not tested, e.g. birds, deer, wild turkeys, pets, wildlife, etc. Additionally, the manure composites were taken from 10 "patties" so it is impossible to know if one in ten or all ten in ten carried the biomarker. If only one in ten carried the biomarker, diluting it with 9 other patties may have reduced the concentration below the method detection limit. When the samples were taken, locations were not randomly selected and the geographic variability was not captured. It is very probable that the bacterial communities of animals living in close proximity to one another will be similar therefore, in order to capture a more representative sample of the watershed, 10 patties should be taken from 10 different farms, not 10 from the same farm (Hartel et. al, 2007, *Geographic sharing of ribotype patterns in enterococcus faecalis for bacteria source tracking*).

24. The "poultry biomarker" MST method is not a standard method nor has it been peer reviewed or third party tested, making it at best a research method and not one that

can or should be used for any regulatory action(s) or used to draw conclusions about the sources of indicator bacteria in the IRW (or anywhere else).

25. The method is based on the amplification of a particular genetic sequence that may or may not be from a live organism. The polymerase chain reaction (PCR) cannot differentiate between live or dead organisms. DNA can persist for long periods of time in the environment, creating positive PCR results long after the indicator or pathogen has been introduced into the environment and has died. Pathogens, in this case salmonella and campylobacter need to be alive to be infectious.

26. The genetic sequence used as the biomarker is a portion of the 16S rRNA gene. The known bacteria that has the closest sequence to this fragment is *Brevibacterium avium*. *Brevibacterium avium* was first identified and isolated from bumble-foot lesions in domestic fowl in 1999. *B. avium* can be cultured and is differentiated from other Brevibaterium species by both its genetic sequence and phenotypical traits e.g., temperature that it grows, utilization of arabinose, etc. It is perplexing that no attempt to culture this "new" bacteria species that the Plaintiff claims to have discovered was carried out. There is no evidence that this "new" organism is viable or pathogenic. Dr. Harwood stated that it is close to the pathogenic, *B. casei*, although genetic analysis indicates that it is much closer to *B. avium*. Being in the same genus as a pathogen does not mean that it will be a pathogen, e.g. the majority of E. coli is nonpathogenic although the nearly identical *E. coli* O157:H7 is highly pathogenic.

27. The Plaintiff claims that the levels of biomarker can be used to quantify the indicator bacteria originating from poultry. However, when developing the quantitative aspect of this assay, negative values were reported for the amount of DNA present. It is impossible to have negative amounts of DNA. Therefore the standard curves used in quantification cannot be correct and the quantitative values reported by the Plaintiff are also incorrect.

28. There is no correlation between the amount of the biomarker and the indicator organism concentration in the litter (see attachment 3). Assuming that all the other technical issues surrounding this new test could be overcome, without a correlation between indicator concentration and biomarker concentration in the litter it is impossible to extrapolate the amount of indicator bacteria originating from poultry in the waters of the IRW.

29. There is no public health linkage between the biomarker and illness rates. No epidemiological studies have been conducted to determine if there is a correlation between these two parameters.

30. Indicator bacteria are ubiquitous and originate from multiple sources in the IRW. Major contributors include cattle, swine and wildlife. The new "biomarker" that the Plaintiffs claim proves that poultry is the primary source of indicator bacteria is not specific therefore, it does not support that conclusion. In addition, there is no correlation between it and the indicator bacteria in the litter so it cannot be used as a quantitative tool. Based on the data that we have reviewed, there is no evidence that

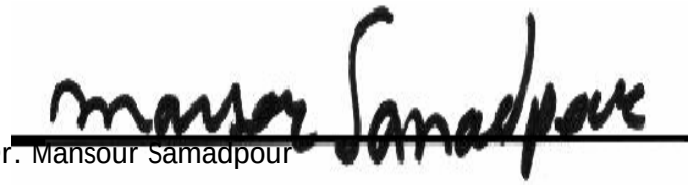
poultry is the major contributor of indicator bacteria or that there is an imminent public health threat in the IRW.

I declare under penalty of perjury that the foregoing is true and correct.

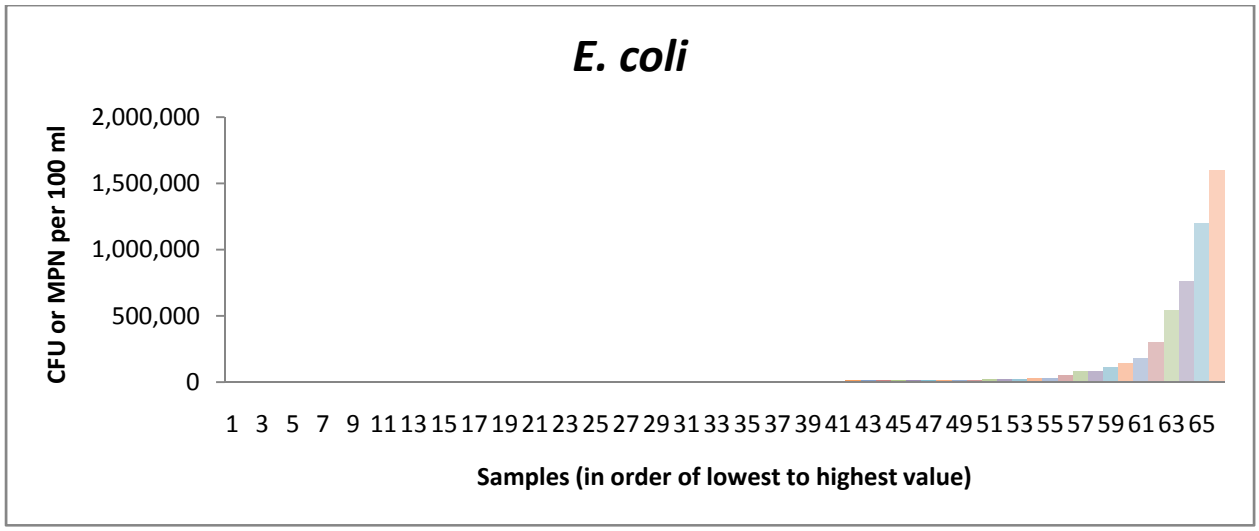
Executed on February 8, 2008

Sam Myoda

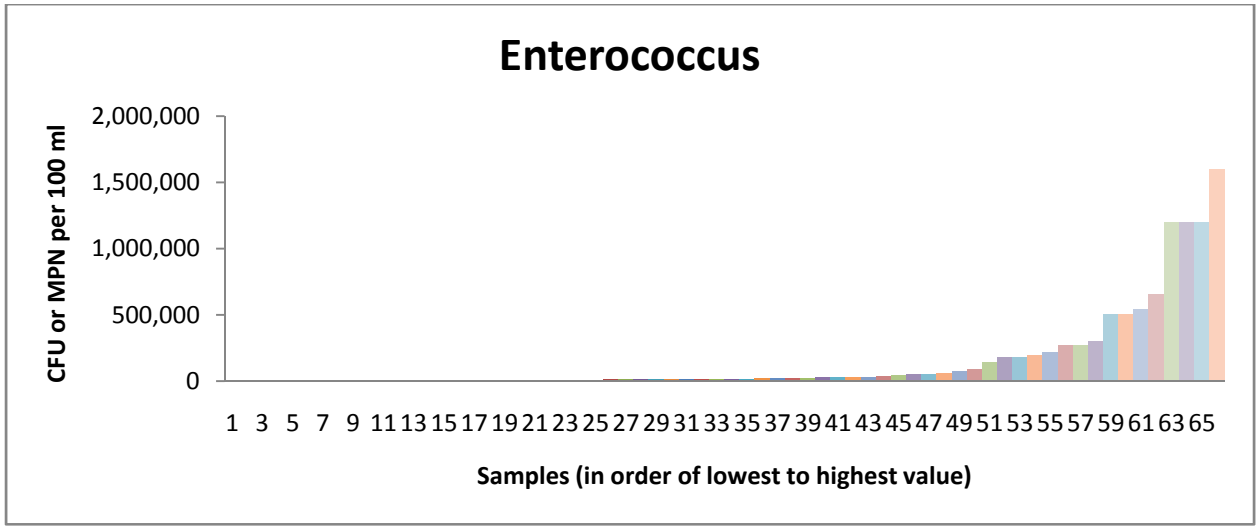
Dr. Samuel P. Myoda



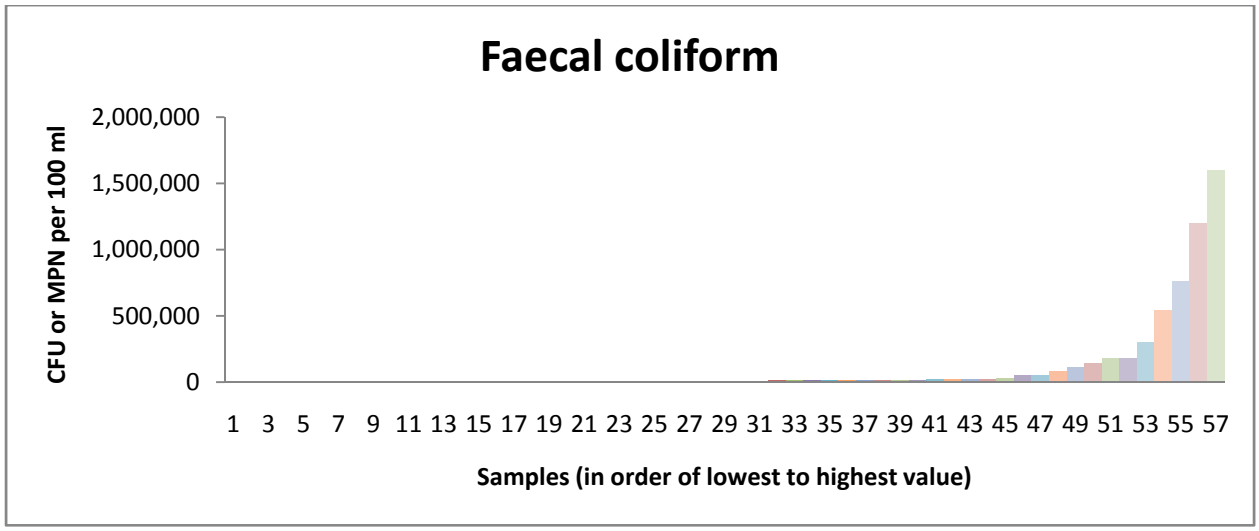
Dr. Mansour Samadpour



Edge of Field	
# of samples	66
	CFU or MPN/100 mL
Geometric mean	4,174
Minimum value	17
Maximum value	1,600,000
80% of all samples below	<24,000
90% of all samples below	<125,000
***Raw sewage ~ 58,000,000 CFU/100mL**	



Edge of Field	
# of samples	66
	CFU or MPN/100 mL
Geometric mean	14,664
Minimum value	110
Maximum value	1,600,000
80% of all samples below	<180,000
90% of all samples below	<503,000

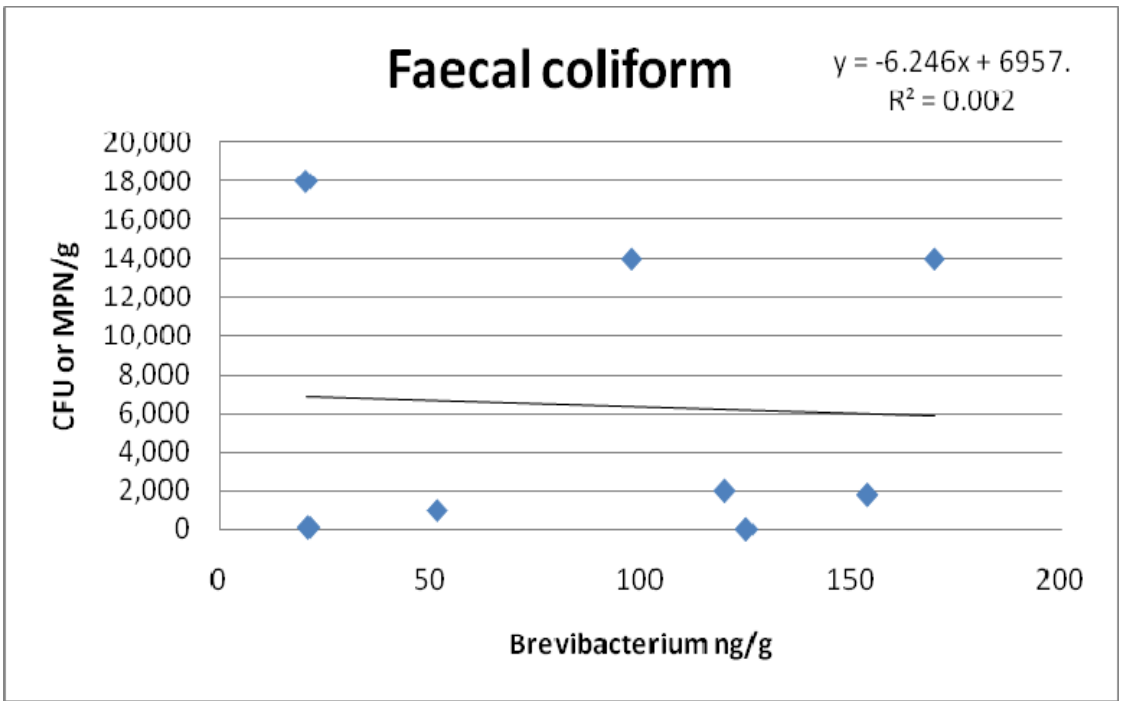


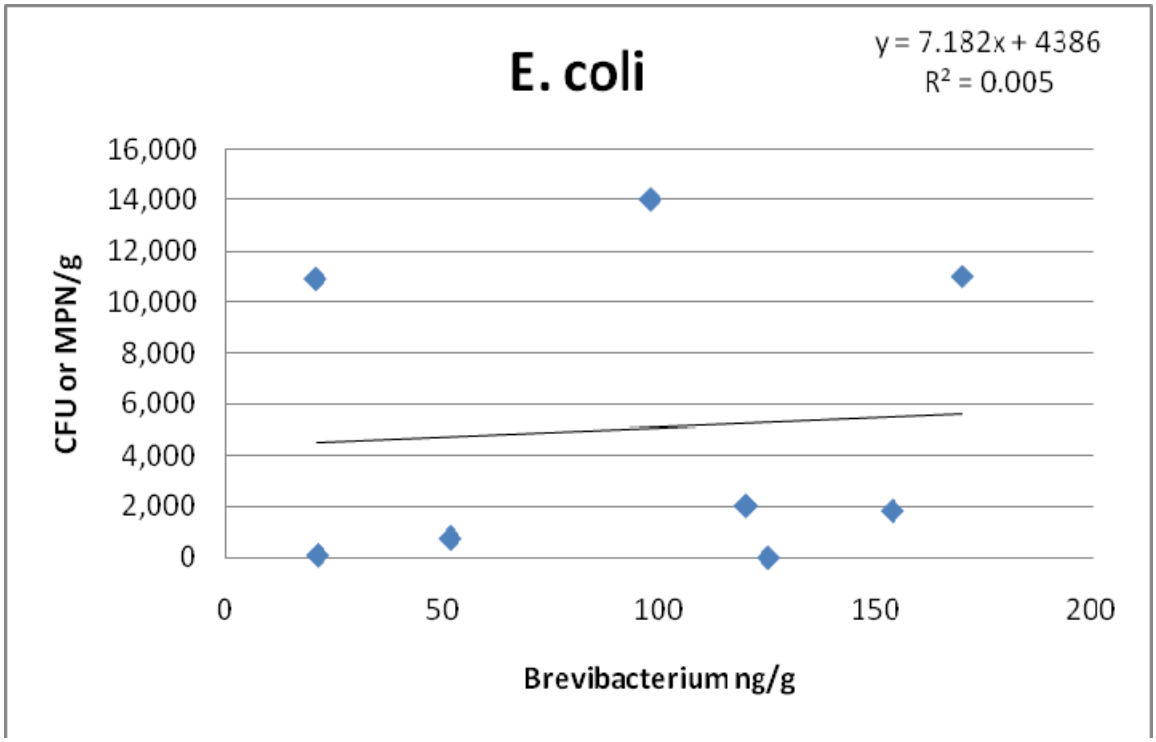
Edge of Field	
# of samples	57
	CFU or MPN/100 mL
Geometric mean	6,371
Minimum value	30
Maximum value	1,600,000
80% of all samples below	<46,000
90% of all samples below	<180,000

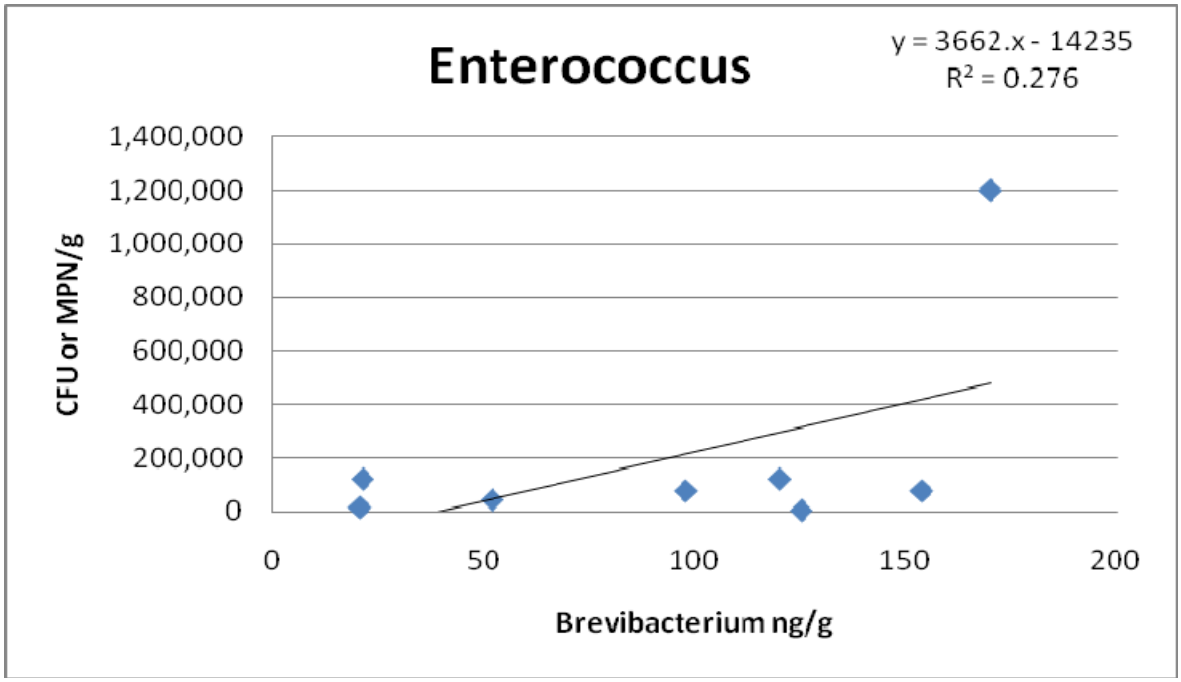
Litter Samples

No correlation between Brevibacterium and indicator organism concentrations

	Faecal coliform MPN or CFU/g	E. coli MPN or CFU/g	Enterococcus MPN or CFU/g	Brevibacterium ng/g
2/2/06	18,029	10,904	13,776	21
7/6/06	94	94	120,000	21.3
6/21/06	980	759	40,988	51.9
8/3/06	14,000	14,000	76,000	98.1
9/22/06	2,000	2,000	120,000	120.1
6/20/06	0.2	0.2	3,800	125.3
8/15/06	1,800	1,800	76,000	154
8/31/06	14,000	11,000	1,200,000	170.1




E. coli
y = 7.182x + 4386
R² = 0.005
16,000
14,000
12,000
10,000
8,000
6,000
4,000
2,000
0
CFU or MPN/g
0
50
100
150
200
Brevibacterium ng/g


Enterococcus
y = 3662.x - 14235
R² = 0.276
1,400,000
1,200,000
1,000,000
800,000
600,000
400,000
200,000
0
CFU or MPN/g
0
50
100
150
200
Brevibacterium ng/g

SAMUEL P. MYODA, Ph.D
IEH Laboratories and Consulting Group
15300 Bothell Way NE
Lake Forest Park, WA 98155
(831) 261-0076

Profile Environmental engineer with extensive regulatory, business and laboratory experience combined with excellent analytical, computer, organizational, public relations, and sales skills. Wide-ranging knowledge of federal and state laws, rules, regulations, policies and procedures including but not limited to monitoring, permitting, compliance and settlements, e.g. Clean Water Act, Clean Air Act, CERCLA, SWDA, RCRA, SARA. Highly motivated and innovational with exceptional project management, training, multitasking and budgeting skills.

Education

University of Delaware, Newark, DE
B.S. Environmental Science (1995), *magna cum laude*
(ranked first in class within major)
Minors in Biology and Economics

University of Delaware, Newark, DE
Ph.D. Civil & Environmental Engineering (2001)

Professional Experience

Institute of Environmental Health/Molecular Epidemiology
Seattle, WA (6/2007 to present)
Vice President

- Expert witness for attorneys on cases such as the September 2006, spinach - *E. coli* O157:H7 outbreak
- Genetic track downs and pathogen detection and quantification
- Laboratory Director and water quality and water systems expert

Delaware Department of Natural Resources and Environmental Control,
Dover, DE (6/2001 to 6/2007, current DWR "Employee of the Year")
Environmental Engineer VI

- Responsible for all aspects of State environmental regulations and standards including authoring, adoption, implementation, compliance and permitting, e.g. TMDL, CAFO, NPDES, MS4, Title V, etc.
- Computer modeling of the fate of air and water pollutants and the effectiveness of reduction strategies, e.g. Watershed Nutrient, DO and pathogen modeling
- Contribute new designs, processes, and techniques, which are regarded as major advances in the field, e.g. established a genetics lab for bacteria source tracking (BST) and served as Director performing BST on all waters in the State of Delaware
- Program management, employee supervision and budgeting (operating and capital)
- Provide technical expertise, direction, guidance and assistance to department personnel, federal/state agencies, industry and private sector organizations
- Formulate long-range objectives, broad policies and strategies and develop and analyze performance measures

- Negotiate and manage consultant contracts, monitor progress and participate in soliciting, reviewing and screening consultants
- Represent the state and department on national and regional committees, e.g. EPA Experts Scientific Workshop on Critical Research Needs for the Development of new or Revised Recreational Water Quality Criteria (3/25-30/2007)
- Plan and implement special studies designed to analyze programs, projects, and services and make appropriate recommendations
- Negotiate settlements, e.g. >$200 million, Title V, air pollution reduction settlement
- Present, prepare and publish reports and documents, e.g. impact statements, technical reports, budget reports, monitoring results, training and safety guidance, QA and QC guidance and peer-reviewed journal articles

University of Delaware, Newark, DE (1997-2001)
Laboratory Manager, Research and Teaching Assistant

- Oversaw all operations within 4 laboratories, duties included technical advisement, scheduling, safety, equipment purchasing and maintenance, ordering and budgeting
- Safety officer
- Assistant to the Department Chairperson
- Taught and trained visiting professors, scholars and graduate and undergraduate students

Lawn Doctor of Newark, Newark, DE (1986-1993, 1995-1997)
Manager

Responsible for the majority of sales and production aspects of operations for three lawn care franchises serving thirty five hundred customers within a 400 sq. mile area. Supervised a staff of fifteen personnel and developed/implemented training programs to maintain multi-state pesticide certification. Interacted with the public on a daily basis.

Selected Accomplishments:

- Set individual and franchise sales records in five of seven years
- Developed IPM (integrated pest management) programs to reduce pesticide usage by 60%
- Spearheaded business expansion into new areas, resulting in the acquisition of a fourth franchise
- Designed employee safety procedures, reducing accidents and chemical exposure
- Expanded sales by initiating a tree and shrub maintenance program and an all-natural lawn maintenance program
- Won “Best Franchise in the Nation” award (1989, 1990, 1992)

Certifications OSHA 40-hour training
Ornamental Horticulture, Longwood Gardens
Delaware Department of Agriculture: categories 03, 5B, 06, 07

Selected Publications and Presentations

Hartel, P.G., **Myoda, S.P.,** Kuntz, R.L., Rodgers, K., Entry, J.A., VerWey, S.A., Schroder, E.C., Calle, J., Lacourt, M., Theis, J.E., Reilly, J.P., Fuhrmann, J.J. (2007), Geographic and Temporal Changes of Enterococcus Faecalis Ribotypes for Bacteria Source Tracking. *Journal of Water and Health (*accepted on 12 Dec 2006)

Samuel P. Myoda, C. Andrew Carson, Jeffry J. Fuhrmann, Byoung-Kwon Hahm, Peter G. Hartel, Helen Yampara-Iquise, LeeAnn Johnson, Robin L. Kuntz, Cindy H. Nakatsu, Michael J. Sadowsky and Mansour Samadpour (2003), Comparison of genotypic-based microbial source tracking methods requiring a host origin database *J Water Health*, 01, pp. 167-180

Jill R. Stewart, R. D. Ellender, Janet A. Gooch, Sunny Jiang, **Samuel P. Myoda** and Stephen B. Weisberg (2003), Recommendations for microbial source tracking: Lessons from a methods comparison study, *J Water Health*, 01, pp. 225-231

Myoda, S.P., Huang, C.P., (2001), A Microscope System with a Dual-band Filter for the Simultaneous Enumeration of *Cryptosporidium parvum* Oocysts and Sporozoites. *Water Research* Vol. 35, No. 17, pp. 4321-4326.

Huang, C.P., Allen, H.A., **Myoda, S.P**., Pirestani, D., Poesponegro, H., Poesponegro, I., Takiyama, L.R., Wang, J. (2000), Chemical Characteristics and Solids Uptake of Heavy Metals in Wastewater Treatment. Water Environmental Research Foundation (WERF), Alexandria, VA.

Huang, C.P., Wang, J., Takiyama, L.R., **Myoda, S.P**. (1998), Fate of Heavy Metals in Wastewater Treatment Systems: Evaluation of Pertinent Parameters and Predictive Models. Workshop Proceedings, 71st WEF Annual Conference and Exposition, WEFTEC'98, Orlando, FL.

Geographic and Temporal Variability of Enterococcus faecalis Ribotypes for Bacterial Source Tracking (ASM – 2003) P. G. Hartel, **S. Myoda**, R. L. Kuntz, K. Rodgers, J. A. Entry, S. A. Ver Wey, E. C. Schroder, M. Lacourt, J. Calle, J. E. Thies, J. P. Reilly, J. J. Fuhrmann

Comparative Diversity of Fecal Bacteria in Agriculturally Significant Animals to Identify Alternative Targets for Microbial Source Tracking (ASM – 2003) J. M. Simpson, **S. Myoda**, D. J. Reasoner, J. W. Santo Domingo

Myoda, S.P., Huang, C.P. (2001) The Ultrasonic Disinfection and Organic Constituent Mineralization of Wastewater. Presenting at the Seventh International Conference on Advanced Oxidation Technologies for Water and Air Remediation, Niagara Falls, Canada (6/28/01)

Laboratories & Consulting Group

ABBREVIATED CURRICULUM VITAE

1. Name: Mansour Samadpour, Ph.D.

2. Position: President
IEH Laboratories & Consulting Group and Molecular Epidemiology, Inc.
Seattle, WA 98155
Off: 206-522-5432, Fax: 206-306-8883, E-mail: ms@iehinc.com

3. Education:
B.S. Microbiology, University of Washington, Seattle, WA, 1981
M.S. Microbiology, University of Washington, Seattle, WA, 1987
Ph.D. Food Microbiology, University of Washington, Seattle, WA, 1990

4. Professional Experience:
President, IEH Laboratories & Consulting Group, 2001-Present
President, Molecular Epidemiology, Inc., 2001-Present
Assistant Professor, Dept. of Environmental Health, University of Washington, 1993-2003
Research Associate, Dept. of Environmental Health, University of Washington, 1991-1993
Post-doctoral Research Associate, Dept. of Environmental Health, University of Washington, 1990-1991
Research Associate, School of Fisheries, University of Washington, 1988-1990
Research Assistant, Dept. of Microbiology, University of Washington, 1978-1980

5. Summary of Publications:

Refereed Journal Articles:	42
Research Reports:	>200
Invited Presentations:	65
Abstracts:	52

6. Membership in Professional Organizations:
American Society for Microbiology
Institute of Food Technologists
International Association for Food Protection
American Society of Animal Science
American Water Works Association
American Public Health Association
Water Environment Federation

7. Selected Activities include:
University of Washington Service: Admissions Committee
Nutritional Sciences Executive Committee
Nutritional Sciences Chair Search Committee
PTAP Committee

Curriculum Committee
Committee on Faculty Responsibilities
Graduate Committee Member

Invited Presentations:

USDA, Manure Management Conference, Des Moines, IA
AMI, Molecular Epidemiology Workshop, Arlington, VA
Tyson Foods, Food Safety Workshop
AMI, Annual Conference
NMI, Annual Convention (Future of Food Microbiology)
AMI, Special Workshop on Epidemiological Approaches in Detection of Foodborne Outbreaks
Annual Food Safety Research Symposium, Recall of Fresh Meat Products

8. Selected Publications in Refereed Journals (1983-2007):

1. Field KG, **Samadpour M.** **"**Fecal source tracking, the indicator paradigm, and managing water quality."Water Res. 2007 Aug;41(16):3517-38. Epub 2007 Jun 27.

2. Stopfort JD, O' Connor R, Lopes M, Kottapalli B, Hill WE, **Samadpour M.** "Validation of individual and multiple-sequential interventions for reduction of microbial populations during processing of poultry carcasses and parts." J Food Prot. 2007 Jun;70(6):1393-401.

3. Meays CL, Broersma K, Nordin R, Mazumder A, **Samadpour M.** " Diurnal variability in concentrations and sources of Escherichia coli in three streams. Can J Microbiol." 2006 Nov; 52 (11):1130-5

4. Meays CL, Broersma K, Nordin R, Mazumder A, **Samadpour M.** " Spatial and annual variability in concentrations and sources of Escherichia coli in multiple watersheds." Environ Sci Technol. 2006 Sep 1; 40(17):5289-96.

5. **Samadpour M**, Barbour MW, Nguyen T, Cao TM, Buck F, Depavia GA, Mazengia E, Yang P, Alfi D, and Stopforth JD. "Occurrence and public health significance of *Listeria monocytogenes*, *Salmonella*, *Escherichia coli* O157:H7 and other shiga toxin-producing *E. coli* in retail fresh ground beef, sprouts and mushrooms." J Food Prot 2006 69:441-443.

6. Lopes M, Stopforth JD, Sucre K, Miksch RR, Giddens E, Reddy MCS, Yemm R, and **Samadpour M**. "Alternative cutting methods to minimize transfer of nervous system tissue during steak preparation from bone-in short loins." J Food Prot 2006 69:220-224.

7. Stopforth JD, Lopes M, Shultz JE, Miksch RR, and **Samadpour M**. "Microbiological status of fresh beef cuts. J Food Prot. 2006 Jun; 69 (6):1456-9.

8. Stopforth JD, Lopes M, Shultz JE, Miksch RR, and **Samadpour M**. "Location of bung bagging during beef slaughter influences the potential for spreading pathogen contamination on beef carcasses." J Food Prot 2006 69:235-238.

9. **Samadpour M**, Roberts MC, Kitts C, Mulugeta W, Alfi D. "The use of ribotyping and antibiotic resistance patterns for identification of host sources of Escherichia coli strains." Lett Appl Microbiol. 2005;40(1):63-8. PMID: 15613004

10. Hooton TM, **Samadpour M. "** Is acute uncomplicated urinary tract infection a foodborne illness, and are animals the source?" Clin Infect Dis. 2005 Jan 15;40(2):258-9. Epub 2004 Dec 22. No abstract available.PMID: 15655744

11. Stoeckel DM, Mathes MV, Hyer KE, Hagedorn C, Kator H, Lukasik J, O' Brien TL, Fenger TW, **Samadpour M**, Stickler KM, Wiggins BA. "Comparison of seven protocols to identify fecal contamination sources using Escherichia coli." Environ Sci Technol. 2004 Nov 15;38(22):6109-17. PMID: 15573614

12. Keene WE, Markum AC, and **Samadpour** M. "Outbreak of *Pseudomonas aeruginosa* infections caused by commercial piercing of upper ear cartilage." JAMA. 2004 291:981-985.

13. Wilkerson C, **Samadpour** M, van Kirk N, and Roberts MC. "Antibiotic resistance and distribution of tetracycline resistance genes in *Escherichia coli* O157:H7 isolates from humans and bovines." Antimicrob Agents Chemother. 2004 48:1066-1067.

14. Myoda SP, Carson CA, Fuhrmann JJ, Hahm BK, Hartel PG, Yampara-Lquise H, Johnson L, Kuntz RL, Nakatsu CH, Sadowsky MJ, **Samadpour M.** "Comparison of genotypic-based microbial source tracking methods requiring a host origin database." J Water Health. 2003 Dec;1(4):167-80. PMID: 15382722

15. Harwood VJ, Wiggins B, Hagedorn C, Ellender RD, Gooch J, Kern J, **Samadpour M**, Chapman AC, Robinson BJ, Thompson BC."Phenotypic library-based microbial source tracking methods: efficacy in the California collaborative study." J Water Health. 2003 Dec;1(4):153-66. PMID: 15382721

16. Davis MA, Hancock DD, Besser TE, Rice DH, Hovde CJ, Digiacomo R, **Samadpour** M, and Call DR. "Correlation between geographic distance and genetic similarity in an international collection of bovine faecal *Escherichia coli* O157:H7 isolates." Epidemiol Infect. 2003 131:923-930.

17. Zhang C, Zhang M, Ju J, Nietfeldt J, Wise J, Terry PM, Olson M, Kachman SD, Wiedmann M, **Samadpour** M, and Benson AK. "Genome diversification in phylogenetic lineages I and II of *Listeria monocytogenes*: identification of segments unique to lineage II populations." J Bacteriol. 2003 185:5573-5584.

18. Davis MA, Hancock DD, Rice DH, Call DR, DiGiacomo R, **Samadpour** M, and Besser TE. "Feedstuffs as a vehicle of cattle exposure to *Escherichia coli* O157:H7 and *Salmonella enterica*." Vet Microbiol. 2003 95:199-210.

19. Renter DG, Sargeant JM, Oberst RD, and **Samadpour** M. "Diversity, frequency, and persistence of *Escherichia coli* O157 strains from range cattle environments." Appl Environ Microbiol. 2003 69:542-547.

20. **Samadpour** M, Kubler M, Buck FC, Depavia GA, Mazengia E, Stewart J, Yang P, and Alfi D. "Prevalence of Shiga toxin-producing *Escherichia coli* in ground beef and cattle feces from King County, Washington." J Food Prot. 2002 65:1322-1325.

21. **Samadpour** M, Stewart J, Steingart K, Addy C, Louderback J, McGinn M, Ellington J, and Newman T. "Laboratory investigation of an *E. coli* O157:H7 outbreak associated with swimming in Battle Ground Lake, Vancouver, Washington." J Environ Health. 2002 64:16-20.

22. Farag AM, Goldstein JN, Woodward DF, and **Samadpour** M**.** "Water quality in three creeks in the backcountry of Grand Teton National Park, USA." Journal Fresh Water Ecology 2001 16:135-143.

23. Hooton TM, Scholes D, Stapleton AE, Roberts PL, Winter C, Gupta K, **Samadpour** M, and Stamm WE. "A prospective study of asymptomatic bacteriuria in sexually active young women." N Engl J Med. 2000 343:992-997.

24. Jackson LA, Keene WE, McAnulty JM, Alexander ER, Diermayer M, Davis MA, Hedberg K, Boase J, Barrett TJ, **Samadpour** M, and Fleming DW. "Where's the beef? The role of cross-contamination in 4 chain restaurant-associated outbreaks of *Escherichia coli* O157:H7 in the Pacific Northwest." Arch Intern Med. 2000 Aug 14-28, 160:2380-2385.

25. *Morbidity and Mortality Weekly Report.* Center for Disease Control. "Outbreak of Salmonella Serotype Muenchen Infections Associated with Unpasteurized Orange Juice -- United States and Canada." MMWR, July 16, 1999 48:577-600.

26. Grimm LM, Goldoft M, Kobayashi J, Lewis JH, Alfi D, Perdichizzi AM, Tarr PI, Ongerth JE, Moseley SL, and **Samadpour** M. "Molecular epidemiology of a fast-food restaurant-associated outbreak of *Escherichia coli* O157:H7 in Washington State." J Clin Microbiol. 1995 33:2155-2158.

27. **Samadpour** M. "Molecular epidemiology of *Escherichia coli* O157:H7 by restriction fragment length polymorphism using Shiga-like toxin genes." J Clin Microbiol. 1995 33:2150-2154.

28. Kim HH, **Samadpour** M, Grimm L, Clausen CR, Besser TE, Baylor M, Kobayashi JM, Neill MA, Schoenknecht FD, and Tarr PI. "Characteristics of antibiotic-resistant *Escherichia coli* O157:H7 in Washington State, 1984-1991." J Infect Dis. 1994 170:1606-1609.

29. **Samadpour** M, Ongerth JE, Liston J, Tran N, Nguyen D, Whittam TS, Wilson RA, and Tarr PI. "Occurrence of Shiga-like toxin-producing *Escherichia coli* in retail fresh seafood, beef, lamb, pork, and poultry from grocery stores in Seattle, Washington." Appl Environ Microbiol. 1994 Mar;60(3):1038-1040.

30. *Morbidity Mortality Weekly Report*. Center for Disease Control. "*Escherichia coli* O157:H7 outbreak linked to commercially distributed dry-cured salami-Washington and California, 1994". MMWR March 10, 1994 44:157-160

31. **Samadpour** M, Grimm LM, Desai B, Alfi D, Ongerth JE, and Tarr PI. "Molecular epidemiology of *Escherichia coli* O157:H7 strains by bacteriophage lambda restriction fragment length polymorphism analysis: application to a multistate foodborne outbreak and a day-care center cluster." J Clin Microbiol. 1993 31:3179-3183.

32. Coyle MB, Carlson LC, Wallis CK, Leonard RB, Raisys VA, Kilburn JO, **Samadpour** M, and Bottger EC. "Laboratory aspects of "Mycobacterium genavense," a proposed species isolated from AIDS patients." J Clin Microbiol. 1992 30:3206-3212.

33. Duguay SJ, Park LK, **Samadpour** M, and Dickhoff WW. "Nucleotide sequence and tissue distribution of three insulin-like growth factor I prohormones in salmon." Mol Endocrinol. 1992 6:1202-1210.

34. Riley DE, **Samadpour** M, and Krieger JN. "Detection of variable DNA repeats in diverse eukaryotic microorganisms by a single set of polymerase chain reaction primers." J Clin Microbiol. 1991 Dec;29(12):2746-51.

35. Stibbs HH, **Samadpour** M, and Ongerth JE. "Identification of *Giardia lamblia*-specific antigens in infected human and gerbil feces by western immunoblotting." J Clin Microbiol. 1990 Oct;28(10):2340-2346.

36. **Samadpour** M, Liston J, Ongerth JE, and Tarr PI. "Evaluation of DNA probes for detection of Shiga-like-toxin-producing *Escherichia coli* in food and calf fecal samples." Appl Environ Microbiol. 1990 56:1212-1215.

37. **Samadpour** M, Moseley SL, and Lory S. "Biotinylated DNA probes for exotoxin A and pilin genes in the differentiation of Pseudomonas aeruginosa strains." J Clin Microbiol. 1988 26:2319-2323.

38. Stibbs HH, **Samadpour** M, and Manning JF. "Enzyme immunoassay for detection of *Giardia lamblia* cyst antigens in formalin-fixed and unfixed human stool." J Clin Microbiol. 1988 26:1665-1669.

39. Moseley SL, Huq I, Alim ARMA, So M, **Samadpour** M, and Falkow S. "Detection of enterotoxigenic *Escherichia coli* by DNA colony hybridization." J Infect Diseases 1980; 142:892-898**.**

40. Moseley SL, **Samadpour** M, and Falkow S. "Plasmid association and nucleotide sequence relationships of two genes encoding heat-stable enterotoxin production in *Escherichia coli* H-10407." J Bacteriol 1983; 186:441-443.